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LABORATORY NOTEBOOK

MBX-1212

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Design primers for sequencing AI/B1 Fusions

1296..2036 ="phbB"

atgac tcagcgcatt gcgtatgtga
 ccggcgcgcatt ggggtggtatc ggaaccgcca tttgccagcg gctggccaag gctggctttc
 gtgtggtggc cgggttcggc cccaactcgc cgcgcgcgca aaagtggctg gacgacgaga
 aggcctctgg cttcgatttc attgcctcgg aaggcaatgt ggcctgactgg gactcgacca
 agaccgcatt cgacaaggtc aagtcgagg tcggcgaggt tgaatgtctg atcaacaacg
 ccggtatcac ccgcgacgtg gtgttcgca agatgaccgc cgcgcactgg gatgcggtga
 tcgacaccaaa cctgactctg ctgttcaacg tcaccaagca ggtgatcgac ggcattggcg
 accgtggctg gggcgcgcat gtcaacatct cgtcgggtga cgggcagaag ggcagttcg
 gccagaccaaa ctactccacc gccaaaggcg gcctgcattg cttcaccatg gcaactggcg
 aggaagtggc gaccaaggcg gtgaccgtca acacggctc tcggggctat atcgccaccg
 acatgggtcaa ggcgactcgc caggacgtgc tcgacaagat gtcgcgacg atcccggtca
 agcgctctgg cctgcccgaag gagatcgct cgatctgcgc ttggtgtctg tcggaggagt
 ccggtttctc gaccggcgcc gacttctcgc tcaacggcgg cctgcatatg ggctga

Table of Contents

40..1221 ="phbA"

a tgactgacgt tgtcatcgta
 tcgcgcgcgc gcaccgcggg cgcaagttt ggcggctcgc tggccaagat ccggcaccg
 gaactgggtg ccgtggtcat caaggccgcg ctggagcgcg ccggcgctcaa gccggagcag
 gtgagcgaag tcatcatggg ccagggtctg accgcgggtt cgggccagaa ccccgacgc
 caggccgcga tcaaggccgg cctgcggcg atggtgcccg ccatgaccat caacaagggt
 tgcggctcgg gcctgaaggc cgtgatgctg gccgccaacg cgatcatggc gggcgacgcc
 gagatcgtgg tggccggggg ccaggaaaac atgagcggc ccccgacgt gctgcccggc
 tcgcgcgatg gtttccgcat gggcgatgcc aagctggctg acaccatgat cgtcgacggc
 ctgtgggacg tgtacaacca gtaccacatg ggcacacccg ccgagaacgt ggccaaggaa
 tacggcatca cagcgaggcg gcaggatgag ttccgcctcg gctcgagaa caaggccgaa
 gccgcgcaga aggcgggcaa gtttgacgaa gagatcgctc ggtgctgat ccgcgacgc
 aaggcgacg cggtggcctt caagaccgac gagttcgtgc gccaggcgcc cagctggac
 agcatgtcgg gectcaagcc cgccttcgac aaggccggca cggtgaccgc ggccaacgcc
 tcgggctga acgacggcgc cgcgcgggtg gtggtgatgt cggcgggcaa ggccaaggaa
 ctgggctga ccccgctggc cagcatcaag agctatgcca acgcgggtgt cgatcccaag
 gtgatgggca tgggcccggg gccggcctcc aagcgcgcc tgtcgcgcgc cgagtggacc
 ccgaagacc tggacctgat agagatcaag gaggccttg ccgcgcaggc gctggcggtg
 eatcagcaga tgggctggga cactccaag gtcaatgtga acggcggcgc catcgccatc
 ggccacccga tcggcgctgc gggtgcctg atcctggtga cgctgctga cgagatgaag
 cgccgtgacg cgaagaaggg cctggcctcg ctgtgcatcg gcggcgccat gggcggtggc
 ctggcagtcg agcgcaata a

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Continued on Page

17

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Luca M. Mulkerson
 Signed

Date

[Signature]
 Signed

Date

Date

AB FUSION CHARACTERIZATION

noticed I had left the MBx240 pTrc AB fusion on the bench for approximately a week.

MBx240 pTrc AB # 10, 11, 12, 13

LB Amp clear / tan no granules

LB Amp white granules
1% glucose

LB Amp white granules
1% glucose
0.1 mM IPTG

eye

light microscopy

start LB glucose O/N culture from freezer stock

50% x = 1% 100 ml

2 hr + 0.5 hr

1/12-16-97 dilute cultures 1:100 into LB 1% glucose Amp

O/N

@ time dil

4 hrs post -

3 hr after inoc.

induction

pTrc-AB 10	0.46335	4.63	0.7839	1.344	1.0204
11	0.55087	5.51	0.7959	1.324	1.0027
12	0.42090	4.21	0.7931	1.513	0.9694
13	0.50844	5.84	0.7476	1.268	0.9897

induced

⊕

⊖

5 mM IPTG

take 300 µl

Samples → 4°C

Continued on Page

92

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Date

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Date

Cont. from p. 88

LOOK @ CULTURES UNDER MICROSCOPE.

DID NOT HAVE TIME WHEN I TOOK
SAMPLES FOR WESTERN.

THEREFORE SAMPLES HAVE BEEN @ RT FROM 5 PM \rightarrow 11:15 AM
* SAMPLES ARE IN LB 1% GLUCOSE *

+ IPTG -

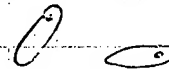
PTC LAB #10

granules
look like peas in
a pod

single granules @ poles



very small



#11

"

"

#12

"

"

#13

"

"

\rightarrow many cells w/ inclusion bodies

Majority of cells in presence 5 mM IPTG
are 2-3x ~~more~~ normal length
while the majority of cells without IPTG
are 1x cell lengths.

of course there is always a small majority
that are 5-10x cell length but only in
IPTG samples

Continued on Page

94

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